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# Non-geographic Variation of *Chaetodipus eremicus* and *Chaetodipus nelsoni* from the Chinati Mountains State Natural Area, Presidio County, Texas, with Comparison to Populations of the Same Species from Brewster County, Texas

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## Abstract

Cranial features were measured and analyzed from a sample of adult pocket mice of the species *Chaetodipus eremicus* and *C. nelsoni* obtained from the Chinati Mountains State Natural Area. A total of 75 specimens was included in the final analyses (38 *C. eremicus* and 37 *C. nelsoni*). Methodologies and statistical tests (MANOVA, Principal Component Analysis, and Discriminant Function Analysis) were conducted in a manner comparable to an earlier study of these two pocket mouse species from Brewster County, Texas, to allow for some comparison of results. Significant intraspecific differences were found between sexes of *C. eremicus* for Greatest Length of Skull measurement (GLS). Also, significant intraspecific differences between sexes were indicated in the Depth of Cranium at Bullae (DCB) character in *C. nelsoni*. Therefore, slight sexual dimorphism was detected within each pocket mouse species. The remaining five cranial characters assessed in this study were significantly different only between species. Within the Principal Component Analysis of the two species, the GLS character explained most of the variation between groups (44.071%). GLS and Cranial Breadth (CB) accounted for more than 50% of variation between species and gender groups. Discriminant Function Analysis revealed some morphometric overlap between species and sexes, indicating the presence of some non-geographic variation within populations of these pocket mice from the Chinati Mountains.

Key words: *Chaetodipus*, Chinati Mountains, *eremicus*, *nelsoni*, non-geographic variation, sexual dimorphism, Texas

## Introduction

The pocket mouse species *Chaetodipus eremicus* (Chihuahuan Desert Pocket Mouse) and *C. nelsoni* (Nelson's Pocket Mouse) are sympatric over many portions of their ranges throughout the Big Bend region of Texas, but usually occupy slightly differing habitat types. *Chaetodipus eremicus* often is found inhabiting areas of sandy soils along washes, creeks, and stream beds, whereas, Nelson's Pocket Mouse seems to prefer rockier habitats and rougher, desert-scrub and sparse grassland habitats (Schmidly and Bradley 2016). Man-

ning et al. (1996) investigated non-geographic variation of these two species of pocket mice from the Rosillas Mountains of Big Bend National Park, Brewster County, Texas, and found significant variation between the two species inhabiting this area but no significant intraspecific differences.

We subsequently obtained specimens of both aforementioned species as the result of a mammalian survey conducted within the Chinati Mountains located in Presidio County, Texas (Jones et al. 2011). Because species and resulting sample sizes from collection efforts were similar to those of the preceding study from Brewster County, we thought it worthwhile to conduct a similar statistical analysis of the two species from Presidio County for sake of comparison of variation between these two sympatric species over a wider portion of their ranges within the Trans-Pecos region of Texas.

Based upon documented range distributions, there appears to be little to no vicariance between and within these two pocket mouse species within this region (Trans-Pecos) of Texas. However, comparisons between localized populations of mammalian species occupying differing montane (and other) areas within their ranges provide additional information regarding the species' natural history and possible environmental effects upon phenotypes. Therefore, the results of our study and comparisons to the Brewster County populations are presented within this manuscript.

## MATERIALS AND METHODS

Specimens of the Chihuahuan Desert Pocket Mouse and Nelson's Pocket Mouse were obtained from study locations within the Chinati Mountains State Natural Area (CMSNA) utilizing Sherman Live Traps baited with rolled oats. Captured mice were subsequently prepared as standard museum skin and skull vouchers and were deposited in the Natural Science Research Laboratory, Museum of Texas Tech University. A sample of approximately equal numbers of males and females of each species (*C. eremicus* and *C. nelsoni*) was considered for our study.

Seven standard cranial measurements were taken on specimens of adult *C. eremicus* and *C. nelsoni* obtained from CMSNA. The measurements included Greatest Length of Skull (GLS), Cranial Breadth (CB), Depth of Cranium at Bullae (DCB), Length of Maxillary Toothrow (MTR), Width of Upper 3rd Molar (WUM3), Greatest Width Upper [maxillary] Tooth Row (WUTR), and Interorbital Breadth (IOB), and follow those described and figured by Wilkins and Schmidly (1979). Zygomatic Breadth (ZB) and Length of Interparietal (LI-I) also were measured; however, a number of the pocket mouse skulls had broken or missing zygomatic elements and subsequent, preliminary testing revealed that interparietal lengths were too variable within our sample to prove useful in subsequent analyses. Therefore, these two cranial characteristics (ZB and LI-I) were excluded from the analysis and results. Additionally, if any individuals were missing particular cranial measurements within the total sample (mostly due to missing or broken cranial elements), those specimens were automatically excluded from all subsequent statistical analyses (MANOVA, Principal Component Analysis (PCA), and Discriminant Function Analysis (DFA)) by the SPSS version 23 software program (2015). Therefore, resultant analyzed sample sizes within and between species and sexes of pocket mice were not equal. Measurements were obtained to the nearest 0.1 mm utilizing digital calipers.

To emulate the work of Manning et al. (1996) for comparison of results, our total sample was partitioned into four groups consisting of species (*eremicus* and *nelsoni*) and gender (female and male) and initially analyzed utilizing the MANOVA tests of SPSS (IBM Corp. 2015). Box's Test and Wilks' tests were performed to test for equality of covariance across groups. Scheffe's test was deemed most appropriate for Post Hoc tests within MANOVA because of the aforementioned unequal sample sizes of the four groups. Scheffe's test is considered to yield conservative results, which reduces the probability of a Type I error of incorrect rejection of a null hypothesis. In addition, our multiple comparisons analyses were evaluated using the Bonferroni correction method, which requires a considerably lower *p*-value in order to assure significance between groups (in our study p-value significance was $p \leq 0.008$). Use of the Bonferroni correction method also reduces the chance of a Type I error.

To further align our work and resultant comparisons to those of Manning et al. (1996), the pocket mouse sample was subjected to the Principle Component Analysis tests of SPSS utilizing the Varimax rotation method. Varimax is the most common rotational method and maximizes the sum of the variances of the squared correlations between the variables and factors. Kaiser-Meyer-Olkin (KMO) tests were conducted to determine whether sample sizes were sufficient for discriminant function analysis, and Bartlett's Test of Sphericity was performed to gauge suitability of resultant responses from discriminant function and principle component analyses. Anti-image correlation testing also was performed to determine validity of each cranial character used in the Principle Component Analysis.

Finally, Discriminant Function Analysis was conducted on the dataset to determine classification power of the analyzed variables (cranial elements). A-priori tests of the dataset for normality, outliers, and non-co-linearity of variables were conducted. Along with other results from the DFA, a scatter-plot diagram was generated to help elucidate the degree in which the analysis factors separated the pocket mouse groups of our study.

## RESULTS

Numbers of each species and gender from our sample, sample means, and standard deviations are presented in Table 1. Sample sizes of each gender/species group remained the same throughout all statistical procedures. Sample sizes of the groups varied because of exclusion of individuals based upon missing cranial measurements. However, despite unequal samples sizes, the means and standard deviations of most of the cranial elements are relatively close when compared between the four sample groups. Standard deviations differ most between groups in GLS, CB, DCB, WUM3, and IOB characters with *C. nelsoni* usually demonstrating the greatest standard deviations within groups (Table 1).

Resultant multivariate analysis of the samples revealed that there were significant differences between some of the subject groups. Consequent results from Scheffe's Test indicated that female and male *C. eremicus* are significantly different in GLS ($p = 0.004$ ). Significant difference in GLS was also inferred between male *C. nelsoni* and female *C. eremicus* ($p = 0.002$), but this is likely due to the larger overall size of *C. nelsoni* when compared to *C. eremicus*. Additionally, males and females of *C. eremicus* differed significantly in CB measurements from male *C. nelsoni* ($p = 0.0005$), as did females of both species when comparing the CB character ($p = 0.001$). Regarding the DC at Bullae cranial character, male *C. nelsoni* individuals were significantly different from all other groups, indicating significant difference between male and female *C. nelsoni* in this cranial characteristic ($p = 0.002$). No significant differences were found between any of the species/sex groups for the MTR cranial character. The only significant difference between the species/sex groups for the WUM3 character was found between male *C. nelsoni* and female *C. eremicus* ($p = 0.0005$). Using the Bonferroni Correction, there was no significant interspecific or intraspecific difference between species/sex groups for the WUTR or IOB cranial characters.

The KMO measure of sampling adequacy for our test was 0.807. As a value of 0.5 is considered to be 'adequate' for Principle Component Analysis of a dataset, our greater KMO value indicated sufficiency of sample sizes. Additionally, the Bartlett's Test of Sphericity was highly significant ($p = 0.0005$), which indicated homogeneity of variances and that PCA analysis upon the dataset would be appropriate (IBM SPSS Support 2018).

It was determined by the Principle Component Analysis that a single character, greatest length of skull (GLS), explained the greatest amount of the variation (44.071%) between sample groups (Table 2). Because a single character accounted for the majority of variation within the sample, no additional rotations were performed in order to account for variation contributions of the remaining characters. Correlations of most of the variables (characters) were not high ($< 0.03$ in most cases); thus, indicating only slight correlation between the analyzed cranial characters.

Table 1. Results of descriptive statistics for MANOVA analysis of *Chaetodipus nelsoni* and *C. eremicus* from the Chinati Mountains, Presidio County, Texas. Cranial measurements (abbreviations are defined in Methods), species and sex, means, standard deviations (SD), and sample size of each species and sex are reported.

| Cranial Measurement | Species and Sex | Mean | SD | N |
|---|---|---|---|---|
| GLS | Female *C. nelsoni* | 25.650 | 0.4872 | 16 |
| | Male *C. nelsoni* | 25.810 | 0.6818 | 21 |
| | Female *C. eremicus* | 25.050 | 0.5621 | 16 |
| | Male *C. eremicus* | 25.755 | 0.6100 | 22 |
| | Total | 25.597 | 0.6549 | 75 |
| CB | Female *C. nelsoni* | 13.144 | 0.4115 | 16 |
| | Male *C. nelsoni* | 13.386 | 0.2851 | 21 |
| | Female *C. eremicus* | 12.650 | 0.1932 | 16 |
| | Male *C. eremicus* | 12.795 | 0.4624 | 22 |
| | Total | 13.004 | 0.4584 | 75 |
| DC at bullae | Female *C. nelsoni* | 8.294 | 0.1237 | 16 |
| | Male *C. nelsoni* | 8.490 | 0.2166 | 21 |
| | Female *C. eremicus* | 8.125 | 0.1065 | 16 |
| | Male *C. eremicus* | 8.182 | 0.1468 | 22 |
| | Total | 8.280 | 0.2118 | 75 |
| Max. TR | Female *C. nelsoni* | 3.319 | 0.1834 | 16 |
| | Male *C. nelsoni* | 3.257 | 0.1434 | 21 |
| | Female *C. eremicus* | 3.244 | 0.1504 | 16 |
| | Male *C. eremicus* | 3.177 | 0.1270 | 22 |
| | Total | 3.244 | 0.1553 | 75 |
| WUM3 | Female *C. nelsoni* | 0.9438 | 0.05123 | 16 |
| | Male *C. nelsoni* | 0.9786 | 0.04586 | 21 |
| | Female *C. eremicus* | 0.9063 | 0.04425 | 16 |
| | Male *C. eremicus* | 0.9545 | 0.05096 | 22 |
| | Total | 0.9487 | 0.05378 | 75 |
| WUTR | Female *C. nelsoni* | 4.363 | 0.1360 | 16 |
| | Male *C. nelsoni* | 4.405 | 0.1284 | 21 |
| | Female *C. eremicus* | 4.306 | 0.1237 | 16 |
| | Male *C. eremicus* | 4.332 | 0.1249 | 22 |
| | Total | 4.353 | 0.1308 | 75 |
| IOB | Female *C. nelsoni* | 6.494 | 0.3172 | 16 |
| | Male *C. nelsoni* | 6.619 | 0.2337 | 21 |
| | Female *C. eremicus* | 6.463 | 0.1928 | 16 |
| | Male *C. eremicus* | 6.555 | 0.1765 | 22 |
| | Total | 6.540 | 0.2348 | 75 |

Table 2. Total variance explained by each component of the Principal Component Analysis. Because a single component explained most variation, the solution was not rotated. Only the GLS component had an Eigenvalue greater than one; however, CB, DC at Bullae, and Max TR each have Eigenvalues close to one.

| Component | Initial Eigenvalues | | | Extraction Sums of Squared Loadings | | |
|---|---|---|---|---|---|---|
| | Total | % of Variance | Cumulative % | Total | % of Variance | Cumulative % |
| 1. GLS | 3.085 | 44.071 | 44.071 | 3.085 | 44.071 | 44.071 |
| 2. CB | 0.966 | 13.800 | 57.871 | | | |
| 3. DCB | 0.878 | 12.549 | 70.420 | | | |
| 4. MTR | 0.812 | 11.601 | 82.021 | | | |
| 5. WUM3 | 0.551 | 7.873 | 89.894 | | | |
| 6. WUTR | 0.436 | 6.223 | 96.117 | | | |
| 7. IOB | 0.272 | 3.883 | 100.000 | | | |

If variation due to CB (13.8%) is added to GLS, over half of the variation (57.871%) within the sample is accounted for (Table 2). Adding variation accounted for by DCB (12.549%) and MTR (11.601%) characters explains 82.021% of the total variation between species and sexes within our sample (Table 2). However, the MTR character was insignificant within the MANOVA analysis; therefore, if only GLS, CB, and DCB are considered, 70.42% of the sample variation is explained by these three cranial characters.

As may be seen in the scree plot (Fig. 1), Eigenvalues of the components clearly show that GLS is most important. A rapid inflection point occurs after

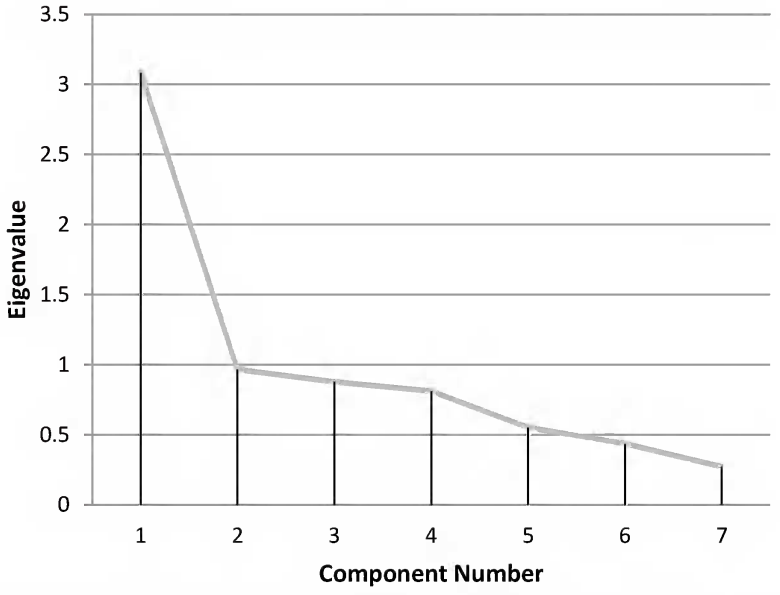


Figure 1. Scree plot of Eigenvalues obtained from the seven cranial components used in PCA analysis of the pocket mice. Component 1 had the only Eigenvalue greater than 1, and a sharp inflection occurred in the scree plot thereafter. Refer to Table 2 for numerical designations and specific Eigenvalues of the individual cranial components.

the first component, indicating that the other variables are of lesser importance, or value, in explaining variation between species and sexes of *C. eremicus* and *C. nelsoni*. This explains why the SPSS analysis only extracted GLS from the data set. Only the GLS character had a calculated Eigenvalue greater than '1', which is the desired condition for principle component extractions. However, as noted above, the addition of the CB and DCB characters help explain slightly greater than 70% of the total sample variation between species and sexes of the pocket mice. Consideration of the MTR character accounts for even more variation; whereas the remaining characters each accounted for less than 10% between groups (Table 2).

Results of a-priori tests for normality of the dataset before our discriminant function analysis revealed that all characters except GLS were significant. Histogram analysis indicated one outlier for the DCB character and two outliers for IOB. However, upon subsequent re-examination of the dataset, we detected no evident disparities in measurements of these cranial elements for the three specimens in question. Also, no co-linearity of variables was detected in the a-priori tests. Therefore, the dataset was left intact for the subsequent DFA.

Box's M-Test was non-significant for the dataset (0.474), indicating homogeneity of co-variance matrices and thus verifying a suitable condition for the dataset. Subsequent Wilk's Lambda tests and standardized canonical discriminant function coefficients indicated that the GLS, CB, and DCB had the most discriminating ability between the variables. Wilk's Lambda values for these three variables were 0.0005, 0.003, and 0.305, respectively. Standardized canonical discriminant function coefficients of the three characters were 0.641, 0.500, and -0.594, respectively. These results were in good agreement with the MANOVA and PCA analyses.

Utilizing the first two functions, 43.8% of *C. nelsoni* females were correctly classified compared to 81% of *C. nelsoni* males (Table 3). The DFA correctly classified 68.8% of female *C. eremicus* specimens and 81.8% of male *C. eremicus* specimens. Overall, 70.7% of the original grouped cases were correctly classified by the discriminate function, and 60% of cross-validated grouped cases were correctly classified (Table 3).

Of the four analyzed groups, female *nelsoni* individuals were misclassified most often, predominantly as males of the same species (Table 3). Female *nelsoni* individuals also were misclassified as either sex of *eremicus*. Male *nelsoni* individuals were misclassified as either females of the same species or male *eremicus* (Table 3). Female *eremicus* individuals were most often misclassified as males of the same species, but also as either male or female *nelsoni*. Finally, male *eremicus* individuals were misclassified as females of both species (Table 3). The resulting scatter-plot illustrates the relationship between groups (Fig. 2). Whereas almost all males of both species separate well, there appears to be much greater overlap between females of both species. However, most females still clustered most closely with their conspecific partners.

Table 3. Discriminant function analysis classification results for *Chaetodipus eremicus* and *C. nelsoni*[a,b]. Correctly classified numbers and percentages are presented in bold text. Original and cross-validated numbers are given along with sample totals.

| | | | Predicted Group Membership | | | | |
|---|---|---|---|---|---|---|---|
| | | Sex | Female *nelsoni* | Male *nelsoni* | Female *eremicus* | Male *eremicus* | Total |
| Original Count | | Female *C. nelsoni* | **7** | 4 | 3 | 2 | 16 |
| | | Male *C. nelsoni* | 2 | **17** | 0 | 2 | 21 |
| | | Female *C.eremicus* | 1 | 1 | **11** | 3 | 16 |
| | | Male *C.eremicus* | 2 | 0 | 2 | **18** | 22 |
| | % | Female *C. nelsoni* | **43.8** | 25.0 | 18.8 | 12.5 | 100.0 |
| | | Male *C. nelsoni* | 9.5 | **81.0** | 0 | 9.5 | 100.0 |
| | | Female *C.eremicus* | 6.3 | 6.3 | **68.8** | 18.8 | 100.0 |
| | | Male *C.eremicus* | 9.1 | 0 | 9.1 | **81.8** | 100.0 |
| Cross-validated Count[c] | | Female *C. nelsoni* | **5** | 5 | 3 | 3 | 16 |
| | | Male *C. nelsoni* | 3 | **15** | 1 | 2 | 21 |
| | | Female *C.eremicus* | 1 | 1 | **10** | 4 | 16 |
| | | Male *C.eremicus* | 3 | 2 | 2 | **15** | 22 |
| | % | Female *C. nelsoni* | **31.3** | 31.3 | 18.8 | 18.8 | 100.0 |
| | | Male *C. nelsoni* | 14.3 | **71.4** | 4.8 | 9.5 | 100.0 |
| | | Female *C.eremicus* | 6.3 | 6.3 | **62.5** | 25.0 | 100.0 |
| | | Male *C.eremicus* | 13.6 | 9.1 | 9.1 | **68.2** | 100.0 |

a. 70.7% of original grouped cases correctly classified.

b. 60.0% of cross-validated grouped cases correctly classified.

c. Cross validation is done only for those cases in the analysis. In cross validation, each case is classified by the functions derived from all cases other than that case.

## DISCUSSION

Most of our findings generally are in good agreement with those of Manning et al. (1996) when comparing non-geographic variation of the Chihuahuan Pocket Mouse and Nelson's Pocket Mouse from the Chinati Mountains, Presidio County, Texas, to those same species from Brewster County, Texas. *Chaetodipus nelsoni* may be distinguished as the larger of the two species in almost every compared characteristic, although there is some degree of overlap.

Unlike the previously studied Brewster County species, there was a slight overall degree of intraspecific variation detected within each species of the Chinati Mountains samples. Sexes of *C. eremicus* were significantly different in the GLS character, whereas *C. nelsoni* sexes were significantly different in the DCB character. Therefore, it would seem that there is some sexual dimorphism, albeit a very small amount, within these two species of pocket mice within the Chinati Mountains region of Texas. Manning et al. (1996) reported no significant intraspecific differences from the Brewster County study. Based upon PCA analysis of the samples, it appears that GLS explains most of the variation in cranial characters, but addition of CB and DC at Bullae measurements help explain greater than 70% of the total variation between sexes and species, and therefore might prove to be useful identification characters in the absence of an entire specimen.

Subsequent discriminant function analysis yielded the same characters as previous MANOVA and PCA analyses as most significantly different and/or important in their explanatory power (GLS, CB, and DCB). However, our DFA results differ somewhat from those

of Manning et al. (1996) as related to their Brewster County, Texas, study. Whereas Manning et al. (1996) achieved 100% correct classification of their species groups, our highest correct classification within the Presidio County specimens was 70.7%. Male centroids were most widely separated, and female centroids of both species were more closely aligned (Fig. 2). This, along with the previous findings of slight sexual dimorphism within each species, would seem to indicate that non-geographic variation is present within the Chinati Mountains populations of *C. eremicus* and *C. nelsoni*.

It is of interest that a small degree of sexual dimorphism was detected in the Chinati Mountains populations of *C. eremicus* and *C. nelsoni*, but not previously within these two species from Brewster County, Texas. Best (1993) studied morphometric variation between and within several species of heteromyid rodents and opined, based upon his own and others' findings, that accurate assessment of sexual dimorphism within a species could not be achieved without an examination of differences at the population level. His statement was based upon discovery of sexual dimorphism (or lack of it) between populations of the same species that were sampled from varying ecological habitats and environmental conditions. Results of this study seem to support these conclusions. Various studies have documented considerable geographic variation in heteromyid populations across their ranges, and it has been noted that the expression of phenotypes is strongly dependent upon ecological factors (Best 1993). Additional studies of these species of pocket mice should be conducted across their ranges within the Trans-Pecos region (and other regions as well) in order to determine the magnitude of geographic and nongeographic variation that exists and possible biogeographical and ecological causes.

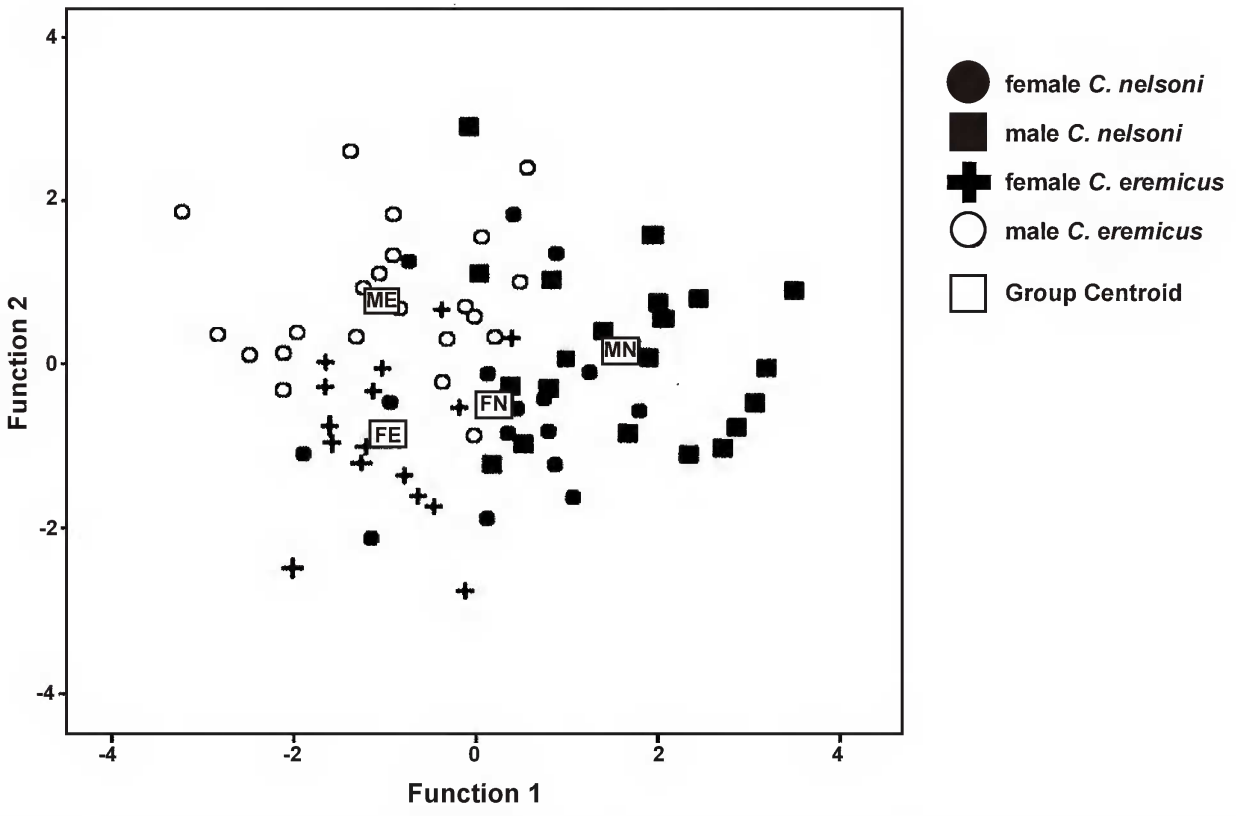


Figure 2. Scatterplot of clustering results from Discriminant Function Analysis of *Chaetodipus eremicus* and *C. nelsoni* species and genders. Male *C. eremicus* and *C. nelsoni* are most widely separated; whereas greater overlap occurs between females of both species.

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